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: 09/804,457

Filed

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March 12, 2001

REMARKS

The Applicants respond below to rejections raised by the Examiner in the Office Action of May 16, 2006.

Rejections under 35 U.S.C. § 103

Claims 1-8, 10-29 and 49-50 stand rejected under 35 U.S.C. § 103(a) as being unpatentable over *Gonzalez et al.* 1995 in view of *Renier et al.* 1995. Applicants respectfully traverse.

Applicants note that the prior art rejections contained in the present Office Action are identical to those originally made over three years ago in an Office Action mailed October 21, 2002. The claims have been modified since then, and the response below is to a large extent a repeat of the response to the October 21, 2002 Office Action, but taking into account the new claim language. It is respectfully submitted that the rejections are overcome for at least the same reasons as set forth in the response to the October 21, 2002 Office Action.

Gonzalez et al.

Gonzalez et al. describe a technique for depolarizing a membrane potential and using a fluorescent dye to monitor both movement across the membrane and cellular transmembrane potential. To effect the electrical potential change in the cell, the reference teaches the use of patch clamping, a technique in which a cell is contacted and then immobilized or "clamped" with a patch pipette. Once the cell is immobilized, a voltage can be applied that will alter the membrane potential of the cell.

Renier et al.

Renier et al. disclose a technique for evaluating the expression of CFTR, a protein associated with cystic fibrosis. The technique relies on fluorescent detection to determine whether a probe moiety has crossed a membrane as an indication of the presence or absence of activated CFTR protein.

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The Prior Art of Record Fails to Teach or Suggest the Claimed Invention:

The prior art of record does not teach or suggest all of the elements of Claim 1 or Claim 21. As all pending dependent claims are based on these two independent claims, the dependent claims are patentable for at least the same reasons.

First, neither Gonzalez et al. nor Renier et al. (either alone or in combination) disclose or suggest all of the elements of Claim 1. Claim 1 recites "[a] method of testing the effect of a candidate compound on the transmembrane potential of one or more biological cells comprising: exposing one or more cells comprising at least one voltage regulated ion channel to said compound; repetitively exposing said one or more cells to a series of two or more electric fields so as to effect a change in transmembrane potential of said one or more cells without using a patch clamp, wherein said transmembrane potential changes predominantly in a single direction away from a starting transmembrane potential over the course of said series of electric fields due to a continuing and additive accumulation of charge in said cell over the course of said series of electric fields; and monitoring, without using a patch clamp, changes in the transmembrane potential of said one or more cells to test the effect of said compound on said one or more biological cells."

Gonzalez et al. do not expose cells to a compound and repetitively expose them to "a series of two or more electric fields so as to effect a change in transmembrane potential of said two or more cells without using a patch clamp." Nor do Gonzalez et al. teach "monitoring, without using a patch clamp, changes in the transmembrane potential of said one or more cells to test the effect of said compound on said one or more biological cells." Gonzalez et al. expose cells to electric fields only to test the performance of the voltage sensitive fluorescent dyes, and does so only with a patch clamp. Further, Applicants note that the Examiner has cited Gonzalez et al. as teaching applying a step potential to neonatal cardiac myocytes to activate voltage gated ion channels. Applicants respectfully submit, however, that such an interpretation mischaracterizes Gonzalez et al. since the myocytes described in the reference were beating spontaneously. See Gonzalez et al. at 1277-78. Accordingly, Gonzalez et al. do not teach using electrical methods to stimulate myocytes. In any case, the cells were not exposed to any compound, so there can be no teaching that the activity of such a compound was monitored. Unlike the embodiments of Gonzalez et al., which use a patch clamp, the embodiments of Claim

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1 monitor changes in transmembrane potential in one or more cells without the use of a patch clamp.

The teachings of *Renier et al.* suffer from similar deficiencies. In *Renier et al.* no electric field is applied at any time, either with or without a patch clamp. Instead, *Renier et al.* teach an evaluation of the expression of functional CTFR by single-cell fluorescence digital imaging using the movement of a probe (DiSBAC₂(3) dye) across the membrane to increase the fluorescence signal. Thus, *Renier et al.* do not teach "exposing one or more cells comprising at least one voltage regulated ion channel to [a] compound [and] repetitively exposing said one or more cells to a series of two or more electric fields so as to effect a change in transmembrane potential." Further, *Renier et al.* does not teach "monitoring, without using a patch clamp, changes in the transmembrane potential of said one or more cells to test the effect of said compound on said one or more biological cells." Thus, *Renier et al.* does not cure the deficiencies of *Gonzales et al.*

Accordingly, neither Gonzalez et al. nor Renier et al., either alone or in combination, teach or suggest "exposing one or more cells comprising at least one voltage regulated ion channel to said compound [and] repetitively exposing said one or more cells to a series of two or more electric fields so as to effect a change in transmembrane potential [or] monitoring, without using a patch clamp, changes in the transmembrane potential of said one or more cells to test the effect of said compound on said one or more biological cells." Because the combination of Gonzalez et al. and Renier et al. does not teach or suggest all of the elements of Claim 1, we respectfully request the Examiner's rejection of Claim 1 (and the claims that depend therefrom) be withdrawn.

Next, the cited prior art does not teach or suggest all of the elements of Claim 21. Claim 21 recites a method of assaying the effect of a compound against a target voltage regulated ion channel, wherein the effect is manifested by transmembrane potential changes. The method comprises "selecting a cell line having a normal resting transmembrane potential corresponding to a selected voltage dependent state of said target voltage regulated ion channel, expressing said target voltage regulated ion channel in a population of cells of said selected cell line, exposing said population of cells to said compound, repetitively exposing said population of cells to a series of two or more electric fields so as to effect a change in transmembrane potential of said population of cells, wherein said transmembrane potential changes predominantly in one

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direction away from a starting transmembrane potential over said course of said series of electric fields due to a continuing and additive accumulation of charge in said cell over said course of said series of electric fields, and monitoring changes in said transmembrane potential of said population of cells to characterize said effect of said compound."

Gonzalez et al. do not teach or suggest expressing a "target voltage regulated ion channel in a population of cells . . . exposing [the] population of cells to [a] compound, repetitively exposing [the] population of cells to a series of two or more electric fields so as to effect a change in transmembrane potential of [the] population of cells . . . [and] monitoring [the] population of cells to characterize [the] effect of [the] compound." In Gonzalez et al., when conducting an experiment in which a potential is applied to a cell, only one cell at a time can be evaluated. This is due in part to the use of patch clamping, a methodology mentioned above in which a single cell is contacted and held in place with a pipette. Prior to Applicants' disclosure, no technique had been found to successfully effect a controlled change in transmembrane potential to a population of cells such that the technique would be suitable for high throughput screening of drug candidate compounds. To the extent patch clamping techniques can be used to control transmembrane potential, these techniques are both too slow and too costly for use in high throughput screening. No other method of applying electric fields has been used for accurate membrane potential control so as to be used in a high throughput assay. Applicants respectfully submit that exposing a population of cells to an electric field is not an obvious variation of exposing one cell to an electric field, particularly where the electric field is able to effect a controlled change in the transmembrane potential of cells.

Further, as indicated above, patch clamping is a technique in which one cell at a time is clamped, stimulated, and measured. By teaching a technique that relies on patch clamping, Gonzalez et al. teach mechanical contact and modification of individual cells. Gonzalez et al. do not teach or suggest exposing more than one cell at a time to an electric field. Thus, Gonzales et al. do not teach "exposing [a] population of cells to a series of two or more electric fields so as to effect a change in transmembrane potential." Renier et al. do not teach the application of any electric field. Without teaching the application of an electric field, Reiner et al. certainly do not teach "exposing [a] population of cells to a series of two or more electric fields so as to effect a change in transmembrane potential of [the] population of cells . . . [and] monitoring [the]

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population of cells to characterize [the] effect of [a] compound." Because neither Gonzales et al., nor Renier et al. teach "exposing [a] population of cells to a series of two or more electric fields so as to effect a change in transmembrane potential," the combination of Renier et al. and Gonzales et al. does not teach all of the limitations of Claim 21. Because the combination of the prior art references do not teach all of the limitations of Claim 21, we respectfully request that the Examiner's rejection of Claim 21 be withdrawn. See M.P.E.P. § 2143.03 (citing In re Royka, 490 F.2d 981 (CCPA 1974)).

CONCLUSION

The Applicants have endeavored to address all of the Examiner's concerns as expressed in the outstanding Office Action. Accordingly, the arguments in support of the patentability of the pending claim set are presented above. In light of these remarks, reconsideration and withdrawal of the outstanding rejections is respectfully requested.

If the Examiner has any questions which may be answered by telephone, he is invited to call the undersigned directly. Please charge any additional fees, including any fees for additional extension of time, or credit overpayment to Deposit Account No. 11-1410.

Respectfully submitted,

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